

$$\frac{1}{\sqrt{2\pi}\sigma_c} \int_{b-2\sigma_s}^{b+2\sigma_s} e^{\left(\frac{-t_c^2}{2\sigma_c^2}\right)} dt = \frac{1}{\sqrt{2\pi}} \int_{\frac{b-2\sigma_s}{\sigma_c}}^{\infty} e^{\left(\frac{-x^2}{2}\right)} dx - \frac{1}{\sqrt{2\pi}} \int_{\frac{b+2\sigma_s}{\sigma_c}}^{\infty} e^{\left(\frac{-x^2}{2}\right)} dx$$

The following calculation steps are required:\*

1.  $2\sigma_s = t_s / \sqrt{2 \ln 2}$
2.  $\sigma_c = t_c / 2\sqrt{2 \ln 2}$
3.  $x_1 = (b - 2\sigma_s) / \sigma_c$
4.  $x_2 = (b + 2\sigma_s) / \sigma_c$
5.  $Q(x_1) = \frac{1}{\sqrt{2\pi}} \int_{x_1}^{\infty} e^{\left(\frac{-x^2}{2}\right)} dx$
6.  $Q(x_2) = \frac{1}{\sqrt{2\pi}} \int_{x_2}^{\infty} e^{\left(\frac{-x^2}{2}\right)} dx$
7.  $I_0 = Q(x_1) - Q(x_2)$
8.  $A_0 = I_0 A_c / A_s$
9. Percentage overlap =  $A_0 \times 100$ ,

where:

$A_s$  = Area of the sample peak of interest determined by electronic integration or by the formula  $A_s = h_s t_s$ .

$A_c$  = Area of the contaminant peak, determined in the same manner as  $A_s$ .

$b$  = Distance on the chromatographic chart that separates the maxima of the two peaks.

$H_s$  = Peak height of the sample compound of interest, measured from the average value of the baseline to the maximum of the curve.

$t_s$  = Width of sample peak of interest at 1/2 peak height.

$t_c$  = Width of the contaminant peak at 1/2 of peak height.

$\sigma_s$  = Standard deviation of the sample compound of interest elution curve.

$\sigma_c$  = Standard deviation of the contaminant elution curve.

$Q(x_1)$  = Integral of the normal distribution function from  $x_1$  to infinity.

$Q(x_2)$  = Integral of the normal distribution function from  $x_2$  to infinity.

$I_0$  = Overlap integral.

$A_0$  = Area overlap fraction.

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\*In most instances,  $Q(x_2)$  is very small and may be neglected.